REMARKS

This amendment is responsive to the non-final Office Action of May 15, 2007. Reconsideration and allowance of claims 1-24 is requested.

Status of the Claims

Claims 1-18 and 22-29 are pending.

Claims 11 and 13 are amended.

Claims 19-21 are cancelled.

The Office Action

Claims 2-4 and 14 stand withdrawn.

Claims 1, 5-13, 15-18, and 22-29 stand rejected under 35 U.S.C. §103(a) as being unpatentable over a combination of:

Prusiner, Novel Proteinaceous Infectious Particles Cause Scrapie, Science. 216(9):136-144 (1982);

Gasset, et al., Perturbation of the Secondary Structure of the Scrapie Prion Protein Under Conditions that Alter Infectivity, PNAS 90:1-5 (1993);

Nandi, et al., Unusual Property of Prion Protein Unfolding in Neutral Salt Solution, Biochemistry 41:11017-11024 (2002);

Cai, et al., Solvent-Dependent Precipitation of Prion Protein, Biochimica et Biopohysica Acta, 1597:28-35 (2002);

Ernst and Race, Comparative Analysis of Scrapie Agent Inactivation Methods, *J. Virol. Methods*, 41:193-202 (1993); and

U.S. Application Serial No. 10/467,591 to Kritzler, et al. (2002).

The Present Application

Prions are resistant to many conventional treatment processes used for destruction of microorganisms. Their behavior also differs in many cases to that of conventional proteins. In particular, conformational changes in the structure of prions in various treatments results in a β-sheet structure which is highly resistant to degradation.

The present inventors have found that a treatment in which one or more phenols is combined with an inorganic salt, e.g., sodium chloride, can inactivate prions on a body.

The References of Record

The Prusiner Article, "Novel Proteinaceous Infectious Particles Cause Scrapie," documents work that proposes that scrapie agent contains a protein which can be inactivated by chaotropic salts such as guanidinium thiocyanate, and can be inactivated by phenol. Chaotropic agents of those that disrupt molecular structure. The results presented in Table 1 on page 138 indicate that scrapie is stable against treatments with ions, such as Na^+, K^+, Cl^-, and SO_4^{2-}. Table 1 also indicates that the scrapie agent is stable in the presence of a list of detergents. These results are repeated on page 139, left column, where the Article indicates that the agent was stable in various ionic and nonionic detergents.

The Prusiner Article notes that "extraction in phenol, a potential denaturant of protein, under various salt and *p*H conditions destroyed infectivity (page 139, left column). However, as is evident from a review of the reference on which this statement is based (S.B. Prusiner, D.F. Groth, S.P.Cochran, F. Marsiarz, M.P. Mc Kinley, H.M. Martinez, Biochemistry 19: p4883 (1980)) (Abstract, attached), extraction with phenol resulted in a finding of virtually no infectivity even after examining such variables as pH, salt concentration, and predigestion of samples with proteinase K, i.e., the effects of extracting with phenol were considered independent of salt concentration.

The Gasset reference discloses that increasing the salt concentrations up to 0.25 M NaCl failed to induce any noticeable change in the amide I′ band of PrP 27-30, indicating no alteration overall in the secondary structure. The conformational stability of PrP 27-30 with varying salt concentrations, the Article suggests, argues for the involvement of multiple molecular forces in the maintenance of amyloid polymers. Thus, it can be concluded that Gasset found that the PrP 27-30 was highly stable in a variety of salt concentrations. Accordingly, Gasset teaches against treatment of PrP 27-30 with salt since it had no effect on the conformational stability.

One of ordinary skill in the art would not be motivated by Gasset to treat a body contaminated with prion with a composition which included an inorganic salt.

Nandi, et al. discloses that the unfolding of cellular prion protein is unaffected by 0.5 molar sodium chloride, as compared to treatment with buffer (FIG. 1, text). Thus one of skill in the art would conclude, based on Nandi, that sodium chloride has no impact on the secondary structure of the prion protein. Also, on page 11020 it is noted that sodium chloride (0.5 M) is without any effect on the thermal

stability of the protein fragment. While the authors note that salt solutions have large effects on the structure and properties of proteins, this is based on studies of proteins in general (p. 11021, left hand column), not on prion proteins. They note that unfolding of prion proteins cannot be explained on the basis of these general considerations (p. 11021, right hand column). Further, they acknowledge that the process of unfolding in prion proteins has not been linked with denaturation of the prion protein (page 1102, right hand column). There is a suggestion on page 11022, bottom of right hand column, that the unfolding of the protein may induce PrP^{Sc} and amyloid formation, thus suggesting a potential for increase in infectivity.

Accordingly, the Nandi reference teaches that the behavior of prion proteins cannot be predicted on the basis of conventional proteins. One skilled in the art, based on Nandi would not be motivated to use an inorganic salt in a composition to treat a prion-infected body.

Cai, et al. reports studies on precipitation of prion protein in various solvents. Precipitation experiments were performed in sodium acetate or sodium phosphate buffers. Sodium chloride and ethyl alcohol were added to reach desired concentrations of 0.05 to 0.025 M and 0-25%, respectively. Salt was found not to effect $PcP^{Sc/RES}$ precipitation in the absence of ethanol, but enhanced the precipitation in the presence of 25% ethanol. The work of Cai demonstrates that the content of the prion residue in the supernatant can be reduced by precipitating the protein. However, this says nothing about the actual infectivity of the prion protein itself, merely its separation from one fraction to another. As noted on page 34, it was only in the presence of ethanol that any effect on precipitation could be determined for salt.

The Ernst and Race reference discloses treating a scrapie-infected hamster brain homogenate with LpH. As mentioned in the Ernst and Race article, LpH is an aqueous acid phenolic disinfectant which contains o-benzyl-p-chlorophenol at 6.1%, as well as p-tertiary amylphenol at 3%, and phenylphenol at 0.5%.

Kritzler, et al. discloses methods for treating a surface suspension or solution contaminated with prion protein with enzymes. In paragraph 41, Kritzler discloses that certain surfactants tend to bind to proteins and initiate unfolding of their tertiary structure. In paragraph 42 it is noted that inorganic salts can induce conformational transitions in proteins. These paragraphs detail the understanding about proteins in general and not about prion proteins. It can be seen from Table 1

that these general assumptions do not apply to prions (as represented by models of proteins such as bovine albumin with high globulin content).

The Claims Distinguish Patentably Over the References of Record

Claim 1 calls for a method of treating a body which is contaminated with prions. The method includes contacting the body with a composition comprising a phenol and a soluble inorganic salt to inactivate prions on the body.

The references cited, alone or in combination, do not suggest such a method.

Prusiner teaches that while scrapie agent is inactivated in chaotropic salts, such as guadinium thiocyanate, it is stable in inorganic ions such as sodium and chloride (Table 1 and page 138, right hand col.). Further, Prusiner teaches that the effect of phenol extraction on infectivity is independent of various salt and pH conditions (page 139, left hand column and reference 24). There is, however, no indication that Prusiner is referring to inorganic salts in this discussion. Accordingly, one of skill in the art would not be motivated to treat a body contaminated with prions with a composition comprising phenol and inorganic salt in view of Prusiner.

The Gasset reference discloses studies on PrP 27-30 polymer, an amyloid form of prion protein formed during enriching fractions for scrapie infectivity which has a high ß-sheet content. Gasset discloses tests on the solubility of PrP 27-30 which includes preparing rods of the material in a phosphate buffer centrifugation, and neutralization of the supernatant which was then normalized to equal salt concentrations. Protein was precipitated with chilled ethanol, as noted on page 2 of the reference. Dispersion of the prion rods into detergent-lipid-protein complexes did not modify the amide I-band line shape, suggesting no effect on the structure. On page 3, right-hand column, the article discusses the changes upon denaturation of PrP 27-30 by SDS. At the bottom of the right-hand column of page 3 it is noted that increasing the salt concentrations up to 0.25 M NaCl failed to induce any noticeable change in the amide I' band indicating no alteration overall in the secondary structure. Higher ionic strengths decreased absorption efficiency which was said to be almost certainly due to poor interaction of the protein sample with the ATR element. The conformational stability of PrP 27-30 with varying salt concentrations, the Article suggests, argues for the involvement of multiple molecular forces in the maintenance

of amyloid polymers. Thus, it can be concluded that Gasset found that the PrP 27-30 was highly stable in a variety of salt concentrations. Accordingly, Gasset teaches against treatment of PrP 27-30 with salt since it had no effect on the conformational stability.

Nandi, et al. discloses the unfolding of cellular prion protein and its refolding to the scrapie isoform. Nandi studied the effects of the various concentrations of sodium sulfate on the CD (circular dichroism) spectra of mouse prion protein, as described on page 11018 and shown in Figure 1. In the discussion which starts at the bottom of the right-hand column on page 11020 of Nandi, the authors note that salt solutions have large effects on the structure and properties of proteins. These statements, however, clearly refer to proteins, in general, not to prions, as evident from the following discussion.

Moreover, Nandi notes that the process of unfolding in prion proteins has not been equated to denaturation (page 11022, right hand paragraph). Indeed, as noted at the bottom of page 11022, right hand column, transition of cellular prion protein to the scrapie isoform is associated with prion unfolding. The authors propose that unfolding of a protein molecule allows intermolecular association through non-covalent interaction would lead to oligomerization and subsequent polymerization to amyloid.

Cai, et al. reports studies on the precipitation of prion protein. Precipitation experiments were performed in sodium acetate or sodium phosphate buffers. Sodium chloride was found not to affect $PcP^{Sc/RES}$ precipitation in the absence of ethanol, but enhanced the precipitation in the presence of 25% ethanol. The work of Cai demonstrates that the content of the prion residue in the supernatant can be reduced by precipitating the protein. However, this says nothing about the actual infectivity of the prion protein itself, merely its separation from one fraction to another. As noted on page 34, it was only in the presence of ethanol that any effect on precipitation could be determined for salt. Thus, Cai provides no motivation for using salt in combination with phenol for treatment of a prion infected body.

Ernst & Race (1993) discloses treating a scrapie-infected hamster brain homogenate with LpH. There is no suggestion in this reference that the composition include a soluble inorganic salt.

Kritzler, et al. discloses methods for treating a surface suspension or solution contaminated with prion protein with enzymes. In paragraph 0041, Kritzler

discloses that certain surfactants tend to bind to proteins and initiate unfolding of their tertiary structure. In paragraph 0042 it is noted that inorganic salts can induce conformational transitions in proteins. These paragraphs detail the understanding about proteins in general and not about prion proteins. Further, as evident from Nandi, conformational changes cannot be assumed to equate with denaturation, but may lead to a conformational change to an infective form. In sum, the references alone or in combination, do not suggest a method for the treatment of a body which is contaminated with prions salts which includes contacting the body with a composition comprising a phenol and a soluble inorganic salt, such as sodium chloride, to inactivate prions on the body. Accordingly, it is submitted that claim 1 and claims 2-9, 14-16, 18, 22, and 25-28 dependent therefrom are patentable over the cited references.

Claim 11 calls for a method of treating a body which is contaminated with prions which includes contacting the body with a composition comprising a phenol and a soluble inorganic salt to inactivate prions on the body, the soluble inorganic salt including sodium chloride.

The references of record do not suggest such a method. In addition to the comments noted above, sodium and chloride ions are taught by Prusiner (Table 1) to be inactive against scrapie agent. There is no suggestion that in the tests using various salts with phenols, that sodium chloride was among the salts tested. Further, Gasset teaches that increasing the salt concentrations up to 0.25 M NaCl failed to induce any noticeable change in the amide I′ band indicating no alteration overall in the secondary structure (at higher concentrations, there was interference with detection procedures which did not allow conclusions to be drawn). Nandi found no effect of salt over and above that of a buffer. On page 11021, left-hand column, it is noted that the chloride ion in the concentration range of 0.1 to 0.7 M has little effect on protein stability.

Accordingly, it is submitted that claim 11, and claims 12 and 17 dependent therefrom, are patentable over the cited references.

Claim 13 calls for a method of treating a body which is contaminated with prions, including contacting the body with a composition comprising a phenol to inactivate prions on the body. The phenol includes *o*-phenylphenol and *o*-benzyl-*p*-chlorophenol in a solution that includes brine.

The references do not suggest such a method. None of the references, with the exception of Ernst and Race, suggests treatment with phenol that includes *o*-phenylphenol and *o*-benzyl-*p*-chlorophenol. There is no suggestion in Ernst and Race that such phenols be used in combination with brine. Moreover, as demonstrated in FIGURE 3F of Cai, salt had no effect on precipitation in an acid solution. Thus, one of ordinary skill in the art would not expect brine to have any effect on an acidic phenol such as *o*-phenylphenol and *o*-benzyl-*p*-chlorophenol.

Accordingly, it is submitted that claim 13 distinguishes patentably over the references of record.

Claim 23 calls for a method of treating a body which is contaminated with prions that includes contacting the body with a composition comprising at least one phenol, the composition comprising a phenol concentration of at least 0.005M and an inorganic salt which is present at a concentration of at least 2% by weight, the phenol including at least one of the group consisting of *p*-chloro-*m*-xylenol; thymol; triclosan; 4-chloro, 3-methylphenol; pentachlorophenol; hexachlorophene; 2,2-methyl-bis(4-chlorophenol); *p*-phenylphenol; 2,3-dimethylphenol; 3,5-dimethoxyphenol; 2,6-dimethoxyphenol; *o*-phenylphenol; *p*-tertiary-amylphenol; *o*-benzyl-*p*-chlorophenol; *p*-chloro, *m*-cresol; *o*-cresol; p-cresol; 2,2-methylenebis(*p*-chlorophenol); 3,4-dihydroxybenzoic acid; *p*-hydroxybenzoic acid; caffeic acid; protocatechuic acid; *p*-nitrophenol; 3-phenolphenol; 2,3-dimethoxyphenol; 2,2-methoxy-bis(4-chloro-phenol); and para-phenylphenol.

The references of record do not suggest treating a body with one or more of the above-mentioned phenols and an inorganic salt at a concentration of at least 2%. Prusiner teaches that the activity of phenol is independent of salt concentration. There is no suggestion that other phenols may benefit from the presence of salt

Accordingly, it is submitted that claim 23 and claim 24 dependent therefrom distinguish patentably and unobviously over the references of record.

Claim 29 calls for a method of treating a body which is contaminated with prions. The method includes contacting the body with a composition to inactivate prions on the body. The composition includes a phenol, a cosolvent, water, and a surfactant selected from the group consisting of sulphonic acids, sulfonates, and combinations thereof.

Prusiner discloses in Table 1 that the scrapie agent is stable in the presence of a list of detergents. These results are repeated on page 139, left column, where the Article indicates that the agent was stable in various ionic and nonionic detergents. In paragraph 0041, Kritzler discloses that certain surfactants tend to bind to proteins and initiate unfolding of their tertiary structure. This paragraph, however, details the understanding about proteins in general and not about prion proteins. Further, as evident from Nandi, conformational changes cannot be assumed to equate with denaturation, but may lead to a conformational change to an infective form.

Thus, it would not be obvious, in view of the cited references, to contact a body with a composition which includes a phenol, a cosolvent, water, and a surfactant selected from the group consisting of sulphonic acids, sulfonates, and combinations thereof.

Accordingly, it is submitted that claim 29 and claim 10 dependent therefrom distinguish over the references of record.

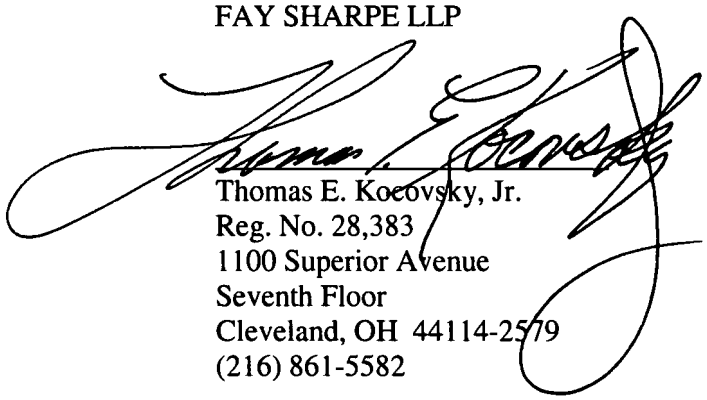
CONCLUSION

For the reasons set forth above, it is submitted that claims 1-18 and 22-29 distinguish patentably over the references of record and meet all statutory requirements. An early allowance of all pending claims is requested.

In the event the Examiner considers personal contact advantageous to the disposition of this case, she is requested to telephone the undersigned at (216) 861-5582.

Respectfully submitted,

FAY SHARPE LLP



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1: Biochemistry. 1980 Oct 14;19(21):4883-91.

Related Articles, Links

Molecular properties, partial purification, and assay by incubation period measurements of the hamster scrapie agent.

Prusiner SB, Groth DF, Cochran SP, Masiarz FR, McKinley MP, Martinez HM.

The scrapie agent causes a progressive degeneration of the central nervous system of animals after a prolonged incubation period. Measurements of incubation period length, defined as the time from inoculation to the onset of clinical signs of neurological dysfunction, were related to the titer of the agent and the dilution of the inoculated sample. Equations defining the relationship provide a new assay for the agent requiring fewer animals than end point titrations. By use of this incubation period assay, the scrapie agent from hamster brain was found to have an s20,w of < 300 S but > 30 S assuming rho p = 1.2 g/cm3. A partially purified fraction P3 was obtained by differential centrifugation and sodium deoxycholate extraction. When P3 was extracted with phenol, virtually no infectivity was found in the aqueous phase even after examining such variables as pH, salt concentration, and predigestion of samples with proteinase K. Nonionic and nondenaturing, anionic detergents

did not inactivate the scrapie agent; in contrast, denaturing detergents inactivated the agent. Sodium dodecyl sulfate (NaDodSO4) inactivated greater than 90% of the agent at a NaDodSO4 to protein ratio of 1.8 g/g. Inactivation by NaDodSO4 appears to be a cooperative process. Addition of a nonionic detergent to form mixed micelles with NaDodSO4 prevented inactivation of the agent by NaDodSO4. Weak chaotropic ions do not inactivate the scrapie agent while strong chaotropic ions like SCN- and Cl3CCOO- destroy infectivity at concentrations of 0.2 M. These data provide evidence in support of a protein component within the scrapie agent which is essential for maintenance of infectivity. Thus, it is unlikely that the scrapie agent is composed only of a "naked" nucleic acid as is the case for the plant viroids.

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